PREPARATION OF METHYL ESTERS BY BORON TRIFLUORIDE METHOD

LOW LAI KIM

INTRODUCTION

Glycerides and phospholipids are saponified, and fatty acids are liberated and esterified in presence of BF_3 catalyst for further analysis by gas liquid chromatography (GLC).

This method is applicable to common animal and vegetable oils and fats, and fatty acids. Unsaponifiables are not removed, and if present in large amounts, may interfere with subsequent analyses.

This method is not suitable for preparation of methyl esters of fatty acids containing major amounts of epoxy, hydroperoxy, formyl, oxo, cyclopropyl, and cyclo-propenyl groups, and conjugated polyunsaturated and acetylenic compounds because of partial or complete destruction of these groups.

APPARATUS

1. Reaction flasks : 50 and 125 ml flasks with outer joints.

2. Condenser : Water-cooled, reflux, with 20 - 30 cm jacket and inner joint.

REAGENTS

1. Boron trifluoride reagent - 125 g BF_3/1 MeOH

   Available commercially as boron trifluoride methanol complex with about 14% BF_3. This reagent is stable for 2 years.

   (Caution : Remove BF3 vapours with effective fume removal device. Avoid contact with skin, eyes, and respiratory tract).

2. Methanolic sodium hydroxide solution (0.5N)

   Dissolve 2 g NaOH in 100 ml MeOH containing < 0.5% H_2O. White precipitate of Na_2CO_3 forming on long standing may be ignored.

3. n-Hexane pure, as determined by GLC.

4. Nitrogen gas containing < 5 mg oxygen/kg.

5. Methyl red solution - 0.1% in 60% ethyl alcohol.

PROCEDURE

Sample Preparation

Precise weighing is not required. Sample size need be known only to determine size of flask and amounts of reagents, according to the following table:

Sample (mg)	Flask (ml)	0.5N NaOH (ml)	BF_3 Reagent (ml)
< 50	10	1 - 2	2
50 - 75	10	2	3
75 - 100	10	3	4
100 - 250	50	4	5
250 - 500	50	6	7
500 - 750	100	8	9
750 - 1000	100	10	12

N.B. Though a 350 mg sample is preferred, it may be difficult to obtain this amount of oil from low fat sample. Hence, 50 - 100 mg sample could be used.

Analytical Procedure

1. Add sample (ca. 350 mg preferred for GLC) to flask and then add 0.5N methanolic NaOH solution and anti-bubbling stone.

2. Attach condenser, and reflux until fat globules disappear (usually 5 - 10 mins at 85°C± 5°C).

3. Add BF_3 solution from bulb or automatic pipette through condenser and continue boiling for 2 min (at 90 - 100°C).

4. Add 2 ml n-hexane through condenser and boil for 1 min.

5. Remove heat, then condenser, and add several ml saturated NaCl solution.

6. Rotate flask gently several times.

7. Add additional saturated NaCl solution to float the n-hexane solution into neck of flask.

8. Transfer about 1 ml upper n-hexane solution into test tube and add small amount anhydrous Na_2SO_4 to remove H_2O. If necessary, dilute solution to concentration of 5 - 10% for GLC.

N.B. BF_3 is very toxic. Work in hood. Wash all glassware immediately after use. If fatty acids containing > 2 double bonds are present, remove air from MeOH and flask by passing in stream of nitrogen gas for a few min. Methyl esters should be analysed as soon as possible. If necessary, n-hexane solution may be kept under N_2 in refrigerator.

REFERENCE

Official methods of analysis of the Association of Official Analytical Chemists (13th Ed.). 1980 : 447.